

Formulation of Nanoemulsion Based Sungkai Leaves Extract (*Peronema canescens* Jack) and Cinnamon Oil (*Cinnamomum Burmannii* (Nees & Th Nees) Blume) as an Antioxidant and Skin Protection Factors (SPF) Agent

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Abstract: Sungkai (*Peronema canescens* Jack) and Cinnamon (*Cinnamomum burmannii*) are herbal plants which are known to have a high content of secondary metabolite compounds as a source of natural antioxidants which are able to inhibit free radical compounds in the body and convert these free radical compounds into non-radical compounds. The greater the free radical scavenger activity in the product, the better the SPF value will be. However, the use of the extract as an active substance in cosmetic dosage forms has a low pharmaceutical elegance, so the extract is formulated using the SNEDDS (Self Nano-Emulsifying Drug Delivery System) technique. Nanoemulsion able to increase the solubility of natural ingredients by eliminating variability in absorption, permeability, and the bioavailability of drugs or bioactive compounds from natural ingredient extracts by maximizing drug penetration into the body and minimizing side effects. The aim of this research is to characterize a nanoemulsion preparation from a combination of sungkai leaves extract and cinnamon in order to obtain the optimal formula for a nanoemulsion preparation as an antioxidant product and skin protection factor agent. This research began with making extracts using the maceration method using 96% ethanol solvent, followed by making nanoemulsions using the spontaneous method. The extract concentration used was 200 mg. Observation of physical properties includes centrifugation test, organoleptic test, percent transmittance, solubility, pH, emulsion type, particle size, polydispersity index, antioxidant activity, SPF value of nanoemulsion preparation and irritation test. Analysis was carried out based on descriptive test parameters. Based on the research results, nanoemulsion preparations were made with Tween 80 surfactant, PEG 400 cosurfactant, VCO oil phase, and aqua deion water phase. So that the optimum formula nanoemulsion preparation (F1) was obtained with an active substance concentration of 100 mg sungkai extract and 100 mg cinnamon extract that met the requirements as a transparent, stable, visual nanoemulsion preparation product, with a transmittance value of 98%, a skin pH range of 6.14, having emulsion type (o/w) with a nanoemulsion particle size of 35.0 nm, PI of 0.479 and has a very strong IC₅₀ antioxidant activity of 20.29 ppm with an SPF value of 7.4 in extra protection.

Keywords: cinnamon, nanoemulsion, sungkai, Skin Protection Factor (SPF)

Abstrak: Sungkai (*Peronema canescens* Jack) dan Kayu Manis (*Cinnamomum burmannii*) merupakan tanaman obat yang diketahui memiliki banyak kandungan senyawa metabolit sekunder sebagai sumber antioksidan alami yang mampu menghambat senyawa radikal bebas dalam tubuh dan mengubah senyawa radikal bebas tersebut menjadi senyawa non radikal. Semakin besar aktivitas penangkap radikal bebas dalam produk, maka nilai SPF akan semakin baik. Namun, penggunaan ekstrak sebagai zat aktif dalam bentuk sediaan kosmetik memiliki keunggulan farmasetika yang rendah, sehingga ekstrak perlu diformulasikan menggunakan teknik SNEDDS (Self Nano-Emulsifying Drug Delivery System). Nanoemulsi mampu meningkatkan kelarutan bahan alam dengan menghilangkan variabilitas absorpsi, permeabilitas, dan bioavailabilitas obat atau senyawa bioaktif dari ekstrak bahan alam dengan memaksimalkan penetrasi obat ke dalam tubuh dan meminimalkan efek samping. Tujuan penelitian ini adalah mengkarakterisasi sediaan nanoemulsi dari kombinasi ekstrak daun sungkai dan kayu manis sehingga diperoleh formula sediaan nanoemulsi yang optimal sebagai produk antioksidan dan skin protection factor. Penelitian ini diawali dengan pembuatan ekstrak menggunakan metode maserasi dengan pelarut etanol 96%, dilanjutkan dengan pembuatan nanoemulsi menggunakan metode spontan (ultrasonikasi). Konsentrasi ekstrak yang digunakan sebesar 200 mg. Pengamatan sifat fisik meliputi uji sentrifugasi, uji organoleptik, persen transmitansi, kelarutan, pH, jenis emulsi, ukuran partikel, indeks

polidispersitas, aktivitas antioksidan, nilai SPF sediaan nanoemulsi dan uji iritasi. Analisis dilakukan berdasarkan parameter uji deskriptif. Berdasarkan hasil penelitian, sediaan nanoemulsi dibuat dengan surfaktan Tween 80, kosurfaktan PEG 400, fase minyak VCO, dan fase air akuades, sehingga diperoleh sediaan nanoemulsi formula optimum (F1) dengan konsentrasi zat aktif ekstrak sungkai 100 mg dan ekstrak kayu manis 100 mg yang memenuhi persyaratan sebagai produk sediaan nanoemulsi yang bersifat transparan, stabil, visual, dengan nilai transmitansi sebesar 98%, rentang pH kulit sebesar 6,14, memiliki tipe emulsi (o/w) dengan ukuran partikel nanoemulsi sebesar 35,0 nm, PI sebesar 0,479 dan memiliki aktivitas antioksidan IC_{50} yang sangat kuat sebesar 20,29 ppm dengan nilai SPF sebesar 7,4 dalam perlindungan ekstrak.

Kata kunci: cinnamon, nanoemulsi, sungkai, Skin Protection Factor (SPF)

INTRODUCTION

Sungkai contains several bioactive compounds with multicomponent interactions in the form of alkaloids, terpenoids, saponins, phenolics, steroids, tannins, and flavonoids in sungkai leaves extract, which can inhibit free radical compounds, and as a source of antioxidants which can inhibit inhibitors of oxidation reactions that occur in the body. The body can increase the synergetic effect through tyrosinase enzyme inhibitors and plant phenolic compounds, inhibiting premature skin ageing (Fadlilaturrahmah *et al.* 2021). When combined with other herbal plants, the compound content in sungkai can produce an excellent synergetic effect. Previous studies carried out by Hamni *et al.* (2022) showed that the combination of methanol extract of Chinese ketepeng leaves (*Cassia alata* L) with six selected plant extracts, including sungkai leaf extract, produces a synergetic plant effect in the form of an additive/indifferent effect. The synergistic effect is an effective way to increase the activity of combined substances compared to the additive effect, where the substance interacts with compounds through a polyherbal combination (Balansa *et al.* 2017).

One of the potential herbal plants that can be combined into sungkai leaf extract as an emulsion preparation and increase the synergetic effect of the compound is a cinnamon extract. The content of bioactive compounds in the form of trans cinnamaldehyde, saponins, catechins, coumarins and tannins in cinnamon extract is a source of natural antioxidant compounds with the ability to capture free radicals or radical scavengers (Watuguly *et al.* 2022). According to research results Antasionasti & Jayanto (2021) that, cinnamon ethanol extract has robust antioxidant activity with an IC_{50} value of 1.939 ± 0.055 μ g/m. Indarto *et al.* (2022) reported that the combination of cinnamon extract (*Cinnamomum burmannii*) and microalgae (*Haematococcus pluvialis*) could be used as a protective preparation for skin protection from sun exposure with the highest SPF value of 12.7 and as an antioxidant compound that inhibits free radicals in the skin. Based on this problem, it is necessary to innovate products from these two natural ingredients to optimize the content of antioxidant compounds as Skin Protection Factor products.

Using these two herbal plants, sungkai and cinnamon, as an emulsion preparation to overcome the problem of bioavailability of natural compound compounds has excellent opportunities but has yet to be widely developed. Furthermore, it can be seen that as many as 40% of plant compounds have low solubility, especially in water (Ramadon & Mun'im 2016), including a combination of sungkai leaf extract and cinnamon. It can also interfere with the body's absorption system because it is difficult for these compounds to dissolve. So, it is essential to use dosage formulation technology because there have been many studies that have only reached the stage of examining extracts from these two plants. Hence, they need to be optimized by carrying out innovative dosage forms, such as nanoemulsions. Nanoemulsion is a drug delivery system with a particle size of 20-200 nm (Pratiwi *et al.* 2018). Nanoemulsion can increase the solubility of natural ingredients by eliminating variability in absorption permeability. It can increase the bioavailability of drugs or bioactive compounds from natural ingredient extracts by maximizing drug penetration into the body and minimizing side effects (Dista *et al.* 2022).

Therefore, a nanoemulsion preparation with a combination of sungkai leaf extract and cinnamon can be an alternative nanoemulsion preparation with functional value for the body. Somehow, there is no research has reported the effect of adding cinnamon to a nanoemulsion preparation of sungkai leaf extract. Thus, this research aims to produce an innovative nanoemulsion combination of sungkai leaf extract and cinnamon as an antioxidant and skin protection factor agent by optimizing the formula. Moreover it can improve the quality of the nanoemulsion preparation by combining sungkai leaf extract and cinnamon as a product rich in antioxidants and a skin protection factor agent to help protect the skin from direct exposure to sunlight.

MATERIALS AND METHODS

Materials

The tools used during this research include a rotary evaporator, UV-Vis spectrophotometry, centrifuge, hot plate, magnetic stirrer (Scilogex), water bath, pH Meter (Digital tester), 100 mL measuring cup, 100 mL erlenmeyer, dropper pipette, tube 10 mL reaction, test tube rack,

vortex, incubator, centrifuge, mask (Softies 3D surgical mask), gloves (latex Minof_ST), sample bottle tube, tweezers, spatula, preparation glass, centrifuge tube and analytical scales (Matrix). The materials used in this research were simplicia sungkai, simplicia cinnamon powder, VCO (Da'I family-lebak), distilled water, and aqua deion. The chemicals used during the research included alcohol 70%, ethanol 96% (Pro Analisa), Tween 80; PEG 400, methanol, DPPH, hexane solution, ethyl acetate, ethanol, and methylene blue (Pro Analisa).

Sample Preparation

The samples used in this research were cinnamon and sungkai leaves (*P. canescens*. Jack), the second and third leaves on fresh tree stalks. Taken from Kedemangan Village, Jambi Luar Kota, Muaro Jambi Regency, Jambi. A total of 7.8 kg of wet sungkai leaves were sorted, cleaned, and washed thoroughly with running water for a short time so that the compounds contained in the sungkai leaves were not lost or dissolved and then drained, separated from the leaf bones and dried for three days at room temperature, in the air and not exposed to direct sunlight. Then, a dry sample of sungkai leaves weighing 2.5 kg was ground using a blender to produce 800 g of sungkai Simplicia, while a dry sample of cinnamon weighing 1 kg was ground using a blender to produce 650 g of cinnamon Simplicia. The drying process aims to ensure that it is durable and can be used for an extended period, does not quickly become mouldy and eliminates enzymatic activity, which can prevent a decrease in quality or damage to the simplicial of sungkai leaves (Handoyo & Pranoto 2020).

Sample Extraction

The extraction method used in this research is maceration, using 96% ethanol solvent, which is soaked for 3 x 24 hr a closed vessel and then concentrated using a rotary evaporator at a temperature of 50°C. Then, the simple yield calculation is carried out from the extract obtained, which aims to determine how much extract is obtained from the simplicial used. The maceration method was chosen in this research because the bioactive compounds in sungkai and cinnamon leaves cannot withstand heating. The extract was mixed with a suitable solvent and then stored at room temperature during storage time. This will minimize damage to the active substance caused by high heating temperatures. Another advantage of this maceration method is that the process does not require high costs and does not require a long time. According to Zubaydah *et al.* (2023), the maceration method is a soaking process using a cold solvent suitable for simplicia, which contains easily soluble secondary metabolites and is not heat resistant like plant compounds. The 96% ethanol solvent used in this research in the maceration method is because

96% ethanol is a polar solvent, easy to obtain, non-toxic and neutral. Besides that, 96% ethanol can extract all low molecular natural substances such as alkaloid compounds, saponins, flavonoids, and other active substances in plant compounds have a relatively low boiling point (78°C) and evaporate quickly, thereby reducing the amount of ethanol carried in the extract.

Maceration was carried out by soaking 517.2 g of sungkai simplicia powder and 518 g of cinnamon Simplicia powder using 96% ethanol solvent separately in the amount of 1000 ml until the Simplicia is wholly submerged and stored in the room for 3 x 24 hr while stirring once in a closed vessel, stirring is carried out. The aim is to evenly distribute the filter fluid so that the concentration will remain maintained due to the difference in concentration between the solvent and the substance being dissolved. After three days, the dregs were squeezed using two layers of filter paper to separate the solution from the dregs and repeated twice. Next, the macerate filtrate was concentrated using a rotary evaporator at a temperature of 40-50°C for approximately 2 hr. This is done to avoid the extract sticking to the round bottom flask of the rotary evaporator; then, the thickening process is carried out using a steam cup placed on top of the water-bath (Larasati & Jusnita 2020).

Characterization Test of Nanoemulsion Preparations

Nanoemulsion Formulation

The nanoemulsion formula combining sungkai leaf and cinnamon extracts was made using the spontaneous method using a magnetic stirrer, which is in line with the research results of Listyorini *et al.* (2018) that (Spontaneous Emulsification) was carried out in order to produce a preparation with a droplet size of 20 - 200 nm. When starting to make an emulsion, mix each formula into a beaker. Mix the oil phase with the extract and stir with a magnetic stirrer for 1 hour, then add the surfactant (Tween 80) and cosurfactant (PEG 400) and pour it into a beaker containing the oil and extract phase slowly and stir with a magnetic stirrer at a speed of 100 – 200 rpm at a hot temperature of 65°C hot plate. The mixture was then sonicated using a bath-type sonicator for 1 hour (Dista *et al.* 2022). Next, the addition of the water phase refers to the research of Listyorini *et al.* (2018) with the ratio (oil phase: deionized water) little by little with light stirring using a magnetic stirrer at a speed of 100 – 200 rpm without heating for 1 hour.

The formula was made with five extract concentration ratios, namely formula 1 (F1) combination, formula 2 (F2) combination, formula 3 (F3) combination, formula 4 (F4) cinnamon single extract and formula 5 (F5) sungkai single extract. Formulation of a nanoemulsion preparation from a combination of sungkai leaf extract and cinnamon is carried out on Table 1.

Table 1. Formulation nanoemulsion

Material	Information	Formulas				
		F1	F2	F3	F4	F5
Cinnamon	Active ingredients (mg)	100*	60*	140*	200*	0
Sungkai	Active ingredients (mg)	100*	140*	60*	0	200*
Tween 80	Surfactant (mL)	20 ml	20 ml	20 ml	20 ml	20 ml
PEG 400	Cosurfact (mL)	10 ml	10 ml	10 ml	10 ml	10 ml
VCO	Oil Phase (mL)	5 ml	5 ml	5 ml	5 ml	5 ml
Aqua Deion	Water Phase (mL)	Add 100	Add 100	Add 100	Add 100	Add 100

(*) in milli gram units

The results of the nanoemulsion preparation were initially evaluated using organoleptic tests, physical stability tests by centrifugation, solubility tests, pH tests and measurement of transmittance values. Further evaluation was carried out on the nanoemulsion selected as the optimal preparation, including testing the particle size and polydispersity index of the optimal nanoemulsion using a Particle Size Analyzer (PSA). After further evaluation, the optimal preparation was tested for antioxidant activity, and the SPF level of the nanoemulsion preparation, and then an irritation test was carried out on the volunteer test group.

Physical Stability Test of Nanoemulsion Preparations

The physical stability test of the nanoemulsion preparation was carried out using a centrifugation test by inserting 2 mL of the nanoemulsion formula preparation into a centrifuge tube and then centrifuging the preparation at 12,000 rpm for 15 min with 1 test measurement. Organoleptic observations of the preparation were carried out, including the colour, aroma and shape, clarity, phase separation and homogeneity of a stable nanoemulsion preparation can be observed with no separation occurring in the two phases (Listyorini *et al.* 2018).

Percent Transmittance and Solubility Nanoemulsion

Observing the visual clarity of the nanoemulsion formula preparation is a qualitative parameter of the dispersion's spontaneity (Shafa & Fitri 2023). According to Redhita *et al.* (2022), the transmittance value will compare the light intensity after interacting with the sample test substance and the light intensity that appears before being involved with the sample test substance. The transmit percentage test aims to ensure the purity of the nanoemulsion preparation as a result of visual observation using UV-Vis spectrophotometry at a wavelength of 650 nm and aqua deion as a comparison or blank. Transmittance values close to 90% - 100% water indicate that the nanoemulsion formulation produces a clear and transparent disperse with nanometer-sized particles (Hastuti & Surkano, 2020; Samsiar *et al.* 2021).

Based on this test, a sample was obtained, which will be continued as the optimal formula for the PSA (Particle Size Analyzer) test for nanoemulsion particle size and polydispersity index.

Solubility Test: was carried out by mixing it in a 10 mL measuring cup. nanoemulsion with organic solvents (1:1) of various levels of polarity, namely hexane, ethyl acetate, ethanol, methanol and water. Each mixture was stirred then the solubility was observed after 6 hr (Jusnita & Syurya 2019).

pH and Nanoemulsion Type

The pH test carried out in this research aims to determine the pH value of the nanoemulsion preparation. It is related to whether the pH of the resulting nanoemulsion preparation meets the safety of the preparation when used and is by the pH on the skin, namely 4.5 – 7. The suitability of the pH value will affect the skin's acceptance of the preparation; if the pH of the preparation is too acidic (smaller than 4, 5), it can irritate the skin, whereas if the pH of the preparation is too alkaline (greater than 7), it will cause a drying effect on the skin. pH testing on nanoemulsion preparations was carried out using a pH meter (Jusnita & Nasution 2019). Before use, the electrode is calibrated or verified using a standard buffer solution of pH 4 and 7. The calibration process is complete when the pH value shown on the screen matches the standard buffer pH value and is stable; after that, the electrode is dipped into the preparation. The pH value of the preparation will be displayed on the screen. pH measurements were conducted at room temperature (20-25°C) (Priani *et al.* 2023). The nanoemulsion type test was carried out by mixing all the nanoemulsion formulations F1, F2, F3, F4 and F5 with methylene blue solution on a glass preparation, then stirring and observing visually. Suppose methylene blue dissolves in the nanoemulsion preparation. In that case, the nanoemulsion is of the oil in water (o/w) type. If the nanoemulsion preparation does not dissolve in methylene blue or clumps on the surface of the nanoemulsion, it is of the water in oil (o/w) nanoemulsion type (Ma'arif *et al.* 2023).

Particle Size Test and Polydispersity Indeks

The particle size test in this study aims to determine whether the nanoemulsion preparation meets the requirements for a nanometer size of <100 nm using PSA (Particle Size Analyzer) as a nanoemulsion preparation product. The working principle of PSA is the scattering of laser light on nanoemulsion preparation particles, which is quickly detected by a photon detector at a certain angle to determine the particle size of the nanoemulsion preparation. The analysis measurement of the nanoemulsion preparation's Polydispersity Index (PI) value aims to show a homogeneous structure or a more stable particle size. A good polydispersity index value is <0.5, while a value >0.5 indicates that the globule distribution is not uniform (Zubaydah *et al.* 2023; Budiarto *et al.* 2020).

Antioxidant Activity Test

The antioxidant activity test of the nanoemulsion preparation in this study was carried out to see the antioxidant activity of the nanoemulsion preparation using the DPPH method as the test solution. According to Firmansyah *et al.* (2022), examining antioxidant activity using the DPPH method is because the process is more straightforward, faster, and more sensitive, and only a small number of test samples are used to evaluate antioxidant activity, so the principle of this method is to measure antioxidant activity quantitatively by measuring radicals. DPPH is captured by the test sample compound, which has antioxidant activity using a UV-Vis spectrophotometric instrument so that the free radical scavenging activity value will be obtained, expressed as the IC₅₀ (Inhibitory Concentration 50%) value.

The DPPH (1,1-diphenyl-2-picrylhydrazil) solution used was made by weighing 2 mg of DPPH, then dissolving it with methanol in a volumetric flask to 100 mL, shaking until homogeneous and obtaining a solution with a concentration of 0.002%. The DPPH solution was stored in a measuring flask covered with aluminium foil to avoid damage due to light. The control blank solution was obtained by adding 2 mL of methanol with 2 mL of 0.002% DPPH solution into a test tube, then vortexing until homogeneous and incubating at room temperature for 30 min in the dark. Nanoemulsion samples from the formulation of nanoemulsion preparations were made into a stock solution of 100 ppm. Dilution was carried out by adding methanol solution so that samples were obtained with test solution concentrations of 10 ppm, 20 ppm, 40 ppm and 80 ppm and 2 ml of DPPH solution was added to each. (0.002%) was vortexed for 2 min. Furthermore, incubate for 30 min Incubation aims to provide an opportunity for substances that act as antioxidants to bind to the DPPH radical compound; then, the absorbance value is read using spectrophotometry at a wavelength of 517 nm. Free radical activity is calculated as the percentage reduction in DPPH

colour. The change in colour of the solution from purple to yellow indicates the efficiency of free radical scavenging. The per cent inhibition curve measured antioxidant activity. Inhibition Concentration (IC₅₀) measures a compound's effectiveness in inhibiting antioxidant activity, which is obtained through the formula equation (1).

$$\% \text{ Inhibition} = \frac{(\text{Abs blanko} - \text{Abs sampel})}{\text{Abs blanko}} \times 100\% \dots (1)$$

Notes: Abs blank: Blank absorbance value; Sample Abs: Sample absorbance value

Based on the immersion percentage value (%inhibition) at each concentration, a regression curve is then created so that the equation $y = bx + a$ is obtained where the sample concentration (ppm) is the abscissa (x - axis) and the immersion percentage value is the ordinate (y-axis) inhibition percentage. Then, the IC₅₀ (Inhibitory concentration) value is calculated, namely the sample concentration with a DPPH absorbance inhibition of 50%. Based on the linear regression equation, the IC₅₀ value will be obtained, where the lower the IC₅₀ value indicates, the higher the antioxidant activity (Cahyaningsih *et al.* 2019).

Nanoemulsion SPF Content/Value Test

The SPF (Sun Protector Factor) value test aims to determine the ability or activity of the sungkai and cinnamon combination extract nanoemulsion to protect the skin. Determination of the SPF value of the nanoemulsion was carried out by taking 1 mL of the nanoemulsion preparation, then dissolving it with methanol in a ratio of 1: 5 mL and homogenizing with a vortex for 1 min. The sample's absorption was measured at a wavelength of 290 - 320 nm using a UV - Vis spectrophotometer (Rahmadani *et al.* 2021). Sample absorbance was recorded every 5 nm interval. The absorption results obtained are then calculated and recorded using the formula (2) (Ningsih & Atiqah 2020).

$$\text{SPF} = CF \times \sum_{290}^{320} \text{EE}(\pi) \times I(\pi) \text{ Abs}(\pi) \dots (2)$$

Notes:

CF : Correction Factor (10)

EE: Spectrum of Erythema Effects caused by UV rays at Wavelengths

I: Intensity spectrum of the sun at wavelengths

Abs: Absorbance of sunscreen products (sample absorption).

Optimal Formula Irritation Test

The irritation test was carried out on the most optimal nanoemulsion formula preparation with the parameters of the nanoemulsion characterization test results, antioxidant results and the SPF value test. The aim is to find out whether the preparation obtained can cause irritation to the skin or not. There

were 10 people as volunteers with three test group categories, namely 4 teenagers (17-22 years), 3 adults (26-35 years) and 3 elderly people (50-60 years) and the test parameters used were observing the level of erythema. (redness) Moreover, degree of edema (swelling) compared to normal skin. By applying the nanoemulsion formula preparation to the lower arm with the results of irritation test observations for 24 hr, 48 hr and 72 hr after administration (Shafa & Fitri 2023).

Data analysis

Data analysis techniques for nanoemulsion characterization were carried out based on the parameters of the Physical Stability Test of Nanoemulsion Preparations, solubility test, nanoemulsion type test, pH test, Percent Transmittance test, Particle Size Test and Polydispersity Index on the optimal formula, as well as an irritation test. Data analysis was carried out using descriptive analysis (Budiarto *et al.* 2020). The DPPH method of antioxidant testing analysis was carried out by looking at the color change of each sample after incubation with DPPH. If all the DPPH electrons pair with the electrons in the test sample, the color of the sample will change from purple to bright yellow. Then the sample's absorbance value was measured using a UV-Vis spectrophotometer at a wavelength of 517 nm, calculating the immersion percentage using a formula based on the concentration immersion percentage and then presented with the results of the regression curve data from the IC50 percentage in the test sample (Cahyaningsih *et al.* 2019).

The SPF value is calculated using a mathematical equation to measure the SPF value using a UV – Vis

spectrophotometer and the sample absorbance is recorded every 5 nm interval. Calculated using the formula (3) (Ningsih & Atiqah 2020)

$$SPF = CF \times \sum_{290}^{320} EE(\pi) \times l(\pi) \text{ Abs}(\pi) \dots (3)$$

RESULTS AND DISCUSSION

Extraction of Sungkai Leaves and Cinnamon

The results of the extraction of sungkai leaf and cinnamon *Simplicia* using the maceration method with 96% ethanol were then concentrated using a rotary evaporator to obtain a thick extract from the *Simplicia*. Then, the yield calculation was carried out to determine how much extract was obtained and the percentage of bioactive content extracted from the *simplicia* used. The results of the extraction process are shown in Table 2 and Table 3.

As for the requirements for a good yield of thick extract if it has a value of > 10%, then it can be seen in Table 3, that the yield value of sungkai extract produces 12.21% and the yield of cinnamon extract is 11.6%, which is more than > 10%. indicates that the extraction process has been carried out properly and optimally. The higher the yield value indicates that the higher the content of substances attracted to a raw material (Setyorini *et al.* 2016).

Nanoemulsion Characterization Test

Nanoemulsion Formulation

The following are the results of making a nanoemulsion formulation from a combination of sungkai leaf extract and cinnamon in Table 4 and Figure 1. The results of making nanoemulsion formulations show that the five nanoemulsion formulations produce clear, transparent, semi-viscous

Table 2. *Simplicia* weighing results

Simplicity	Initial Weight	Dry Weight	Pollinated	After sifting
Sungkai Leaves	7.8 Kg	2.5 Kg	800 g	600 g
Cinnamon	-	1 Kg	850 g	650 g

Table 3. Rendement results

Extract	Initial weight (g)	Weight of extract (g)	Yield (%)
Sungkai Leaves	517.2	63.2	12.21
Cinnamon	518	60.5	11.67

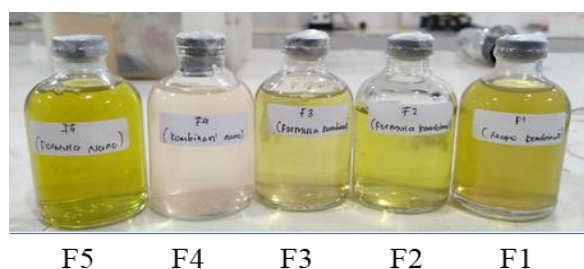


Figure 1. Nanoemulsion preparation

Table 4. Nanoemulsion formulation

Formulas	Test Parameters		
	Color	Aroma	Clarity
Formula 1 (F1)	Pale yellowish green	Distinctive Smell	Clear
Formula 2 (F2)	Pale yellowish green	Distinctive Smell	Clear
Formula 3 (F3)	Pale yellowish green	Distinctive Smell	Clear
Formula 4 (F4)	Clear light brownish	Distinctive Smell	Clear
Formula 5 (F5)	Yellowish green	Distinctive Smell	Clear

preparations with a distinctive aroma that is not different organoleptically. However, the colour parameters show that the combination extract concentration has a different colour with a more yellow-green colour in sequence from formulas F5, F1, F2, and F3. For F4, it has a typical clear colour from the cinnamon extract. Meanwhile, it can be seen that the yellow colour produced from the nanoemulsion prepared comes from Tween-80 as a surfactant. This research uses sungkai leaves and cinnamon extracts as active substances for making nanoemulsion formulations.

The selection of sungkai leaves and cinnamon as active substances is based on the content of bioactive compounds in the two plants. According to Fadlilaturrahmah *et al.* (2021), The tannin and flavonoid content in sungkai leaf extract can inhibit free radical compounds, which are known to have activity as natural antioxidants, increase the synergetic effect in the form of tyrosinase enzyme inhibitors, and the phenolic compounds in sungkai leaves can inhibit premature ageing of the skin. The same content in cinnamon, according to Watuguly *et al.* (2022), can also overcome the problem of premature skin ageing and inhibit free radical compounds that occur in the body. Where is the bioactive content in the form of trans cinnamaldehyde, saponins, catechins, coumarins and tannins in the cinnamon extract is a source of natural antioxidants capable of acting as radical scavengers.

Tween 80 was used as a hydrophilic non-ionic surfactant in this research because it increased the clarity of the physical appearance of the nanoemulsion and was considered a material that was not toxic and irritated the skin when used. PEG 400, a cosurfactant, can help accelerate nanoemulsion formation and increase active substances through increased penetration. This finding is in line with that conducted by Shabrina & Khansa (2022); Nursal *et al.* (2019) stated that the use of Tween-80 combined with PEG 400 was able to increase the stability of the preparation, the physical appearance of the nanoemulsion could indicate the homogeneity of the preparation, opacity, clarity and thermodynamic size of the oil phase droplets used. This is also to the research results of Zubaydah *et al.* 2023; Priani *et al.* 2023, using Tween 80 as a surfactant can produce stable nanoemulsions and emulsify oil by adsorbing on the surface of oil globules so that it forms a single

layer and reduces the tension between oil and water, Tween 80 as a surfactant because it has a low level of irritation, is not easily affected by pH and is biocompatible. The use of PEG 400 as a co-surfactant is a mid-chain hydrocarbon that can help the solubility of solutes and stabilize the layer on the globules in nanoemulsion preparations so that the globule diameter becomes more constant and can be maintained. According to Issusilaningtyas & Indratmoko (2021), Tween 80 and PEG 400 were chosen in preparations for making nanoemulsions because they have HLB (Hydrophile lipophile balance) values as required in self-nanoemulsifying drug delivery system (SNEDDS) preparations ranging from 15 – 21.

The oil phase used in preparing nanoemulsions plays a key role as a carrier that can dissolve lipophilic active substances. The oil phase used in this research is VCO (Virgin Coconut Oil), which is pure coconut oil widely used in nanoemulsion preparations from previous research. Yuliani *et al.* (2016) reported that the use of VCO as the oil phase in making nanoemulsion preparations has several advantages, especially the VCO content in the form of long-chain triglyceride (LCT), which can produce nanoemulsion preparations that are more stable, transparent and clear compared to using oil. contains medium chain triglycerides (MCT). The LCT content is more soluble in water because there are more polar groups than the oil phase with MCT content, so the LCT oil phase carrier material is better at interacting. It can expand the surface and form a very stable emulsion from making nanoemulsion preparations.

Physical Stability Test of Nanoemulsion Preparations

The results of the physical stability tests carried out in this research showed that the five formula preparations made were F1, F2, F3 with varying concentrations of combination extract nanoemulsions and F4, F5 were nanoemulsion preparations with a single concentration of sungkai and cinnamon extracts obtained from the centrifugation test of the preparations. nanoemulsion shows that all nanoemulsion formula preparations do not experience changes in color, distinctive aroma, liquid form, the clarity of the preparation remains stable, and there is no separation, precipitation or turbidity from the initial form of the preparation. These results indicate

that the nanoemulsion formulation has good physical stability (Table 5).

The physical stability test of the nanoemulsion preparation using a centrifugation test was carried out to see whether phase separation occurred in the nanoemulsion preparation. The results obtained are in accordance with research conducted by Listyorini *et al.* (2018) which stated that the centrifugation test was carried out to determine whether or not the nanoemulsion formula preparation had phase separation that occurred due to the earth's gravitational force within a period of one year and whether the resulting nanoemulsion preparation clear color, stable and there is no separation then the preparation can be said to be a stable and good nanoemulsion preparation. In line with research results from Redhita *et al.* (2022); Nurhidayati *et al.*, (2020), reported that testing using centrifugation can describe stability and determine the impact of product transportation shocks on the physical appearance of a product. In accordance with the research results of Zubaydah *et al.* (2023), the occurrence of phase separation in the centrifugation test is due to the ability of surfactants and cosurfactants to reduce the surface tension of the emulsion between the oil phase and the unstable water phase and can affect the phase separation in the nanoemulsion preparation.

Percent Transmittance Test

The following are the results of the percent transmittance test for nanoemulsion preparations. Based on the test results for the percentage of transmittance produced from nanoemulsion preparations, it was found that all preparations had a stable, clear and transparent visual appearance with a value of 98% - 99%. The five formula preparations for F1, F2, and F3 are known to be nanoemulsion preparation formulas from a combination of sungkai

and cinnamon extracts which have transmittance values that are not much different from the results of nanoemulsion preparations with a single extract concentration of F4 and F5 so that the results of the transmittance values obtained show that The combination of extracts given in the nanoemulsion preparation does not affect the results of the transmittance value and has met the requirement level for the percent transmittance of the nanoemulsion preparation of 90% - 100%, the same as the water phase value (Table 6).

The percent transmittance test carried out is in line with the results of previous research conducted by Beandrade (2018); nanoemulsion with a transmittance value > 90% is a clear, transparent and stable nanoemulsion preparation. In line with the research results of Listyorini *et al.* (2018) with aqua deion as a blank or comparison because aqua deion does not have particles that can withstand light transmission, so it will transmit light that passes through it without any light scattering effects, therefore the per cent transmittance value of aqua deion 100%. Apart from that, the composition of the surfactant and cosurfactant used can also influence the results of the nanoemulsion transmittance per cent test, by what was done by Shabrina *et al.* (2020); Widayastuti & Saryanti (2023) reported that the use of Tween 80 and PEG 400 as surfactants and cosurfactants with various concentrations will produce a per cent value of transmit approaching 100%, which means the preparation has clear and transparent physical characteristics. The transmittance value, which is influenced by the concentration of Tween 80 through the mechanism of reducing the surface tension between the oil and water phases in the nanoemulsion system, will produce a smaller particle size. The higher the clarity

Table 5. Physical stability Nanoemulsion

Formulas	Color	Aroma and shape	Clarity	Separation Phase	Homogeneity
F1	Pale Yellowish Green	Typical/Liquid	Clear	Not Separating	Homogeneous
F2	Pale yellowish green	Typical/Liquid	Clear	Not Separating	Homogeneous
F3	Pale yellowish green	Typical/Liquid	Clear	Not Separating	Homogeneous
F4	Light brownish clear	Typical/Liquid	Clear	Not Separating	Homogeneous
F5	Yellowish Green	Typical/Liquid	Clear	Not Separating	Homogeneous

Table 6. Percent transmittance test results

Formulas	Percent Transmittance (%)
F1	98
F2	99
F3	99
F4	98
F5	99

value, the better the nanoemulsion preparation. The amount of surfactant and cosurfactant used in this research with a composition greater than that of the oil is to be able to cover the oil droplets when they emulsify in water and produce droplet sizes in the nanometer range from the results of the nanoemulsion preparation transmit value test for (F1) with a total combined extract concentration of 100 mg of sungkai and 100 mg cinnamon with a transmit value of 98% was taken as the optimal formula for further tests using a PSA (particle size analyzer) to determine the particle size of the nanoemulsion preparation.

Solubility Test

The solubility of nanoemulsion preparation products is an important factor in maximizing their utilization. Table 7 shows the results of the solubility test for the nanoemulsion preparation.

Based on the results of the solubility test in this study, the aim was to compare the solubility of nanoemulsion preparations from various levels of polarity of the test solvent. From the test results, it is known that the five nanoemulsion preparations that were made were completely soluble and homogeneous, with the observation characteristic of being transparent white in ethanol, methanol and distilled water, and insoluble in hexane and ethyl acetate solvents, where the nanoemulsion preparation produced two immiscible phases. The results of this research are in line with the results of research conducted by Jusnita & Nasution (2019) and Larasati & Jusnita (2020) that the nanoemulsion preparation cannot be dissolved in ethyl acetate, hexane and is completely soluble in ethanol, methanol and distilled water solvents. According to Jusnita & Syurya

(2019), the insolubility of nanoemulsion preparations in hexane and ethyl acetate solvents is because the extract used mostly consists of polar water, while the hexane solvent is non-polar. This is supported by Jusnita *et al.* (2019), who stated that the solubility of a substance in a solvent is largely determined by the suitability of the properties between the dissolved test substances, namely the like dissolves like nature, partly due to its polarity. However, this research did not test the polarity value of the resulting nanoemulsion, so the results show no change in the properties of the extract concentration when it has become a nanoemulsion preparation. The small size of the nanoemulsion preparation has a larger surface area, so it can help increase the solubility of the preparation. Nanoemulsion in solvents.

Nanoemulsion Type and pH

The results of the pH measurement test for each nanoemulsion preparation formula show that the preparation has a pH value that is within the skin's pH range. The average pH value of the experiment results is between 6.07 – 6.25, which shows that each preparation formula is good for use on the skin. It can be seen that the results of the formulation of nanoemulsion preparations with combination extracts in the F1, F2 and F3 formula preparations show a pH value that is not much different from the formula preparations with single extracts F4 and F5 so that the combination extract preparation formula is classified as good for use in the skin pH range and does not cause The pH value results are smaller (<4) than the single extract preparation formula or the results show a higher pH value (>7). The following are the results of the pH test for the nanoemulsion preparation in Table 8.

Table 7. Solubility test results of nanoemulsion preparations

Solvent Type	Solubility				
	F1	F2	F3	F4	F5
Hexane	-	-	-	-	-
Ethyl Acetate	-	-	-	-	-
Ethanol	+	+	+	+	+
Methanol	+	+	+	+	+

Notes: (-): Not dissolved/ (+): Dissolved

Table 8. Nanoemulsion pH Test Results

Formulation Nanoemulsion	pH			pH Average
	I	II	III	
F1	6.02	6.13	6.27	6.14
F2	6.19	6.25	6.30	6.25
F3	6.10	6.14	6.19	6.14
F4	6.05	6.07	6.08	6.07
F5	6.06	6.19	6.20	6.15

According to Ma'arif *et al.* (2023) several factors can influence the stability of inappropriate pH values of nanoemulsion formulas, namely the ratio of the concentration of the formula and the type of extract used, the type of surfactant, cosurfactant and other excipients. Based on research results reported by Avianka *et al.* (2022); Samsiar *et al.* (2021) one of the requirements for sunscreen preparations is to have an SPF (Sun Protection Factor) protection value for the skin, where the pH value of the preparation has a value range of the skin's pH. Products that have a pH that is too high or too low will irritate the skin, a pH value below 4.5 can irritate the skin, while a pH value above 6.5 will cause scaly skin. The results of the emulsion type test for the nanoemulsion preparation presented in Table 9.

The emulsion type test results show that the five nanoemulsion formula preparations that were made (F1, F2, F3, F4, and F5) have an oil-in-water (o/w) emulsion type. It can be seen from the test results that the methylene blue dye dissolves completely, producing a blue color. which is homogeneous and there is no formation of lumps from the test results. The results of this research are in line with the research results of Dista *et al.* (2022) which used a nanoemulsion formula composition with sungkai leaf extract as the active ingredient, Tween 80 surfactant, PEG 400 cosurfactant with a VCO oil phase having an oil-in-water (o/w) emulsion type. and does not show separation because the addition of Tween 80 and PEG 400 will reduce the surface tension of VCO with aqua deion so that phase separation does not occur (Figure 2).

Meanwhile, it is known that the use of Tween 80 and PEG 400 in this research has an HLB (Hydrophilic lipophile balance) value which supports

the formulation of nanoemulsion preparations to form oil-in-water (o/w) type emulsions. Rahmadevi *et al.* (2020) and Ma'arif *et al.* (2023) reported that using methylene blue as a solvent in a nanoemulsion type test, basically methylene blue is polar, so that when dissolved with a nanoemulsion preparation the methylene blue dye will dissolve and diffuses evenly throughout the o/w nanoemulsion preparation because hydrophilic or polar components dominate the nanoemulsion preparation. Budiarto *et al.* (2020) and Lestario *et al.* (2024) reported that the results of the nanoemulsion type test produced a water-in-oil type (o/m), so the particles of methylene blue dye would experience changes and there would be a clustered structure on the surface of the media.

According to Zubaydah *et al.* (2023); Fayakun & Prihantini (2023) several factors can influence the results of the oil-in-water nanoemulsion type test (o/w or (o/w)), especially influenced by the Hydrophilic lipophile balance (HLB) value, if the HLB value is in the range of 8 – 18 So the nanoemulsion formed is of the oil-in-water type. The HLB value as the polarity value of the use of oil, noionic surfactants and cosurfactants, describes the balance between hydrophilic and lipophilic groups, so it tends to form oil-in-water emulsions. Meanwhile, nanoemulsion preparations with the (o/w) oil-in-water type, Kusumawardani *et al.* (2020) reported that this type of oil-in-water emulsion has several advantages, especially helping to increase solubility, absorption and bioavailability. And if applied in skin preparations, it will give a soft feeling to the skin, is easy to wash, is long-lasting and in drug-releasing preparations is good when used on the skin and increases easy absorption into the skin tissue.

Table 9. Nanoemulsion type test results

Formulas	Solubility of Methylene Blue	
	Late	Clot
Formula 1	+	-
Formula 2	+	-
Formula 3	+	-
Formula 4	+	-
Formula 5	+	-

Information: (+): Dissolved/ (-): Clumping

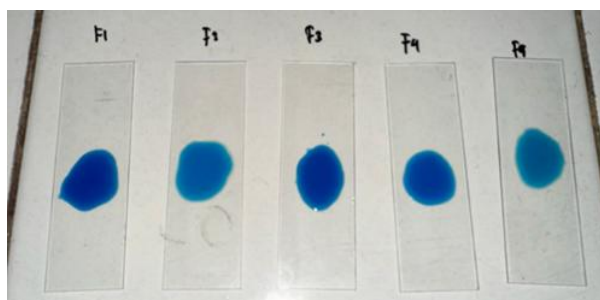


Figure 2. Emulsion type test results

Particle Size Test and Polydispersity Index

In this test, the nanoemulsion formulation formula (1) (F1) was determined with a combined concentration of extracts (100 mg sungkai and 100 mg cinnamon) as the optimal formula based on the results of further tests (transmittance, antioxidants and SPF levels in the nanoemulsion preparation). The following are the results of the particle size test and polydispersity index of the nanoemulsion preparation (F1) in Table 10.

Based on the results obtained for the nanoemulsion formula 1 (F1) particle size, namely 35.0 nm and the droplet polydispersity index (PI) value of 0.479, these results show that the particle size of the F1 nanoemulsion preparation is <100 nm with a polydispersity index (PI) value, nanoemulsion preparation is less than 1. The PI value in Table 10 means that the nanoemulsion preparation has a fairly good level of droplet size distribution uniformity; from these results, the F1 nanoemulsion preparation has fulfilled the requirements as a nanoemulsion preparation product. The result is in line with the research results of Budiarto *et al.* (2020), Nurdianti *et al.* (2022), Issusilaningtyas & Indratmoko (2021) nanoemulsion which has a size of <100 nm and a polydispersity index (PI) value of fewer than 1 shows result in accordance with previous transmittance value results which provide an initial picture of the size of the nanoemulsion particles so that the preparation indicates a homogeneous size distribution. Beandrade (2018) stated that a uniform particle size distribution will cause the active substance to be absorbed at a relatively equal and fast rate, thereby increasing its bioavailability. Dista *et al.* (2022) reported that the polydispersity index (PI) value in nanoemulsion shows the quality of homogeneity and stability of the droplet size of the preparation. The smaller the PI value approaches 0, the more homogeneous the nanoemulsion particle

size will be. In accordance with the research results of Fayakun & Prihantini (2023), the polydispersity index value will indicate the particle size distribution, which provides information about the physical stability of a dispersion system by increasing the value. A low polydispersity index value indicates that the dispersed system formed will be stable in the long term. Meanwhile, what can influence the particle size distribution in nanoemulsion preparations is the energy used during the process of making nanoemulsion preparations, such as the stirring and heating processes, which can influence the resulting particle size distribution. The results of particle size testing of nanoemulsion preparations carried out are in line with the research results of Jusnita & Nasution (2019) that the rotation speed used in the formulation process will influence the results of nanoemulsions with an average particle size of less than 100 nm, this is in line with Shabrina's research, (2022) that nanoemulsions with surfactant Tween 80 and cosurfactant PEG 400 will produce particle sizes of less than 50 nm.

One of the factors that can influence the particle size results in nanoemulsion preparations is the results of research conducted by Rizki *et al.* (2023) who reported that the influence of the type of surfactant used is based on the hydrophilic-lipophilic balance value HLB (Hydrophilic lipophilic balance). Nonionic surfactants (have no charge) have an affinity close to the water and oil phases, will reduce surface tension, produce a small droplet charge without any influence on sample oxidation, are safe, biocompatible and are not influenced by the pH of the medium.

Antioxidant Activity Test

The following are the results of the antioxidant activity test for nanoemulsion preparations in Figure 4. Combined graph of antioxidant activity.

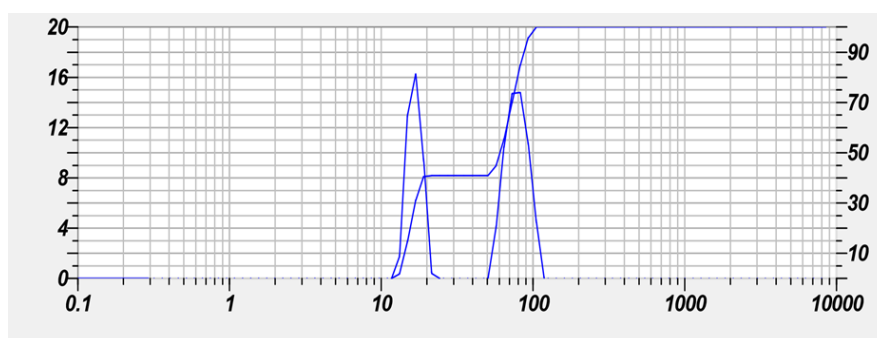


Figure 3. Particle size and polydispersity index of nanoemulsion

Table 10. Particle size and polydispersity index test results

Formula	Particle Size (nm)	Particle Size Distribution
F1 (Combination of 100mg sungkai+ 100 mg cinnamon)	35.0	0.479

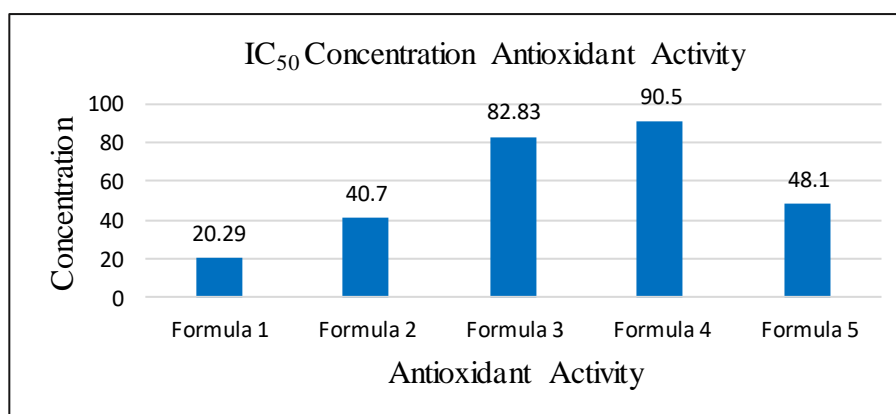


Figure 4. Composite graph of antioxidant activity

The antioxidant activity was carried out by using a DPPH radical agent. The result showed that there was a decrease in the absorbance value of the sample with the value of the sample concentration level in each nanoemulsion formula preparation tested, namely F1, F2, F3, F4 and F5. Then, in the test process, the colour change occurred in the test sample of the nanoemulsion preparation after administering the DPPH solution, ranging from purple to yellow. In accordance with the research results of Junista & Syurya (2019), Larasati & Jusnita (2020), and Pratiwi *et al.* (2018), there was a change in the colour of the test sample solution during the process of examining antioxidant activity due to the reduction of DPPH radical compounds by antioxidants, which indicates the efficiency of preventing free radicals. Meanwhile, there is an influence that the higher the concentration of the test sample, the more antioxidant compound particles it contains and the higher the antioxidant activity in it, so the absorbance value of the test sample will decrease with the concentration level used when measuring antioxidant activity, and the increase in per cent inhibition is influenced by the decrease. Absorbance value produced by the test sample: The decrease in absorbance value is caused by the high concentration of the test sample. This results in a higher sample concentration, a smaller absorbance value, and a higher percent inhibition.

The results of calculating the IC₅₀ value obtained from the linear regression equation of the standard curve of the relationship between the concentration of the nanoemulsion formula preparation and determined by calculations using the equation $y = a x + b$ where the value of $y=50$ and the x value shows the IC₅₀ of the nanoemulsion formula preparation. The IC₅₀ values were obtained respectively. 20.29 ppm in formula F1, 40.7 ppm in formula F2, 80.69 ppm in formula F3, 90.5 ppm in formula F4, and 48.1 ppm in formula F5, which can be seen from the results of the combined graph of antioxidant activity in Figure 5. If compared to the IC₅₀ value of the combined extract nanoemulsion formula preparation with the IC₅₀ results of the single extract nanoemulsion formula preparation, it is known that

the IC₅₀ value of formulas F1 and F2 has a higher per cent inhibition compared to the single extract formula preparation of F4 with the results not being too different for the F5 preparation. Meanwhile, the IC₅₀ results for F3 show per cent inhibition results that are not much different from F4, which has strong antioxidant properties. From the results of examining the antioxidant activity of the nanoemulsion formula preparation in this research, it is known that the preparation of the nanoemulsion formula with a combination extract does not affect the results of the IC₅₀ value of the preparation and shows that the combination of the two plant extracts used as active substances, namely sungkai and cinnamon in the nanoemulsion preparation, has activity properties. Very strong antioxidants for F1 and F2, while F3 has strong antioxidant properties with a comparison of the results of the antioxidant activity properties of the single extract formula preparation, F4 has strong antioxidant properties, and F5 has very strong antioxidant properties.

The five nanoemulsion preparations that were made had antioxidant properties according to their strength category with an IC₅₀ value of less than 50 ppm, including very strong antioxidants, and it was found to be a strong antioxidant compound if the IC₅₀ was between 50 ppm - 100 ppm, then if the IC₅₀ value was in the range 101 ppm - 150 ppm, then it is considered to be in the medium category. Meanwhile, if the IC₅₀ is between 150 ppm - 200 ppm, the compound is considered to have very weak antioxidant activity (Taufik *et al.* 2021). Apart from that, the results of the antioxidant activity in the nanoemulsion formula combined with sungkai and cinnamon extracts show that the antioxidant compounds contained in the two plant extracts are not damaged or reduced after being made into a dosage product. This is in line with the research results of Larasati & Jusnita (2020), which state that the turmeric antioxidant compounds were not damaged in making turmeric extract nanoemulsion preparations after they were made into nanoemulsion preparations.

The differences in antioxidant properties obtained in this study are known to be due to the comparison of the concentration levels of the combination extracts used in the nanoemulsion preparation. When compared with previous research from the results of Magfirah's research (2021), the reason for the difference in the antioxidant activity of nanoemulsions using parang romang leaf extract is that the content in the extract is still in the form of complex compounds (not pure). Meanwhile, according to Jusnita & Syurya (2019), the results of their research provide a comparison of extract concentration levels in the nanoemulsion preparation of Moringa leaf extract, which will influence the results of antioxidant activity. By adding more extract to the nanoemulsion formulation, the higher the antioxidant activity obtained in nanoemulsion preparations, as well as the treatment of characterization tests on nanoemulsion preparations, including nanoemulsion particle size tests and transmit test results, have also been described as having good antioxidant activity results. From this statement, it can be concluded that the results of this research in making nanoemulsion preparations with a combination of sungkai and cinnamon extracts produce similarities with the results of previous research conducted by Jusnita & Syurya (2019); Magfirah (2021) it is known that in making nanoemulsions using active ingredients from plant extracts with different concentrations in the nanoemulsion formulation, it will affect the results of the antioxidant activity values of the nanoemulsion preparations and the active substances used have also gone through a long process before becoming thick extracts and as nanoemulsion preparation products.

Besides that, the incubation treatment factor in the antioxidant activity test can also influence the results of the IC_{50} of the nanoemulsion preparation. It can be seen from this research that the nanoemulsion formula preparation with a combination extract in F3 and the single extract nanoemulsion preparation F4 during the antioxidant activity test experienced a storage period that was not long enough for the incubation period with the other formulas because at

the time of testing the preparation was not sufficient for cuvettes in UV-Vis at during the test, it isn't easy to show repeated absorbance values, but the antioxidant activity can still be read with the antioxidant activity results being classified as strong with IC_{50} values > 100 ppm. In accordance with the research results of Shafa & Fitri (2023), the incubation period during the DPPH test is not long enough so that the extract contained in the emulsion has not reacted completely with free radicals and will affect the antioxidant activity of the nanoemulsion preparation.

Nanoemulsion SPF Content/Value Test

The SPF (Sun Protection Factor) value test in this research was carried out to ensure that the nanoemulsion preparation of sungkai and cinnamon combination extract could absorb UV radiation that hit the skin. the test results for the levels/SPF values of nanoemulsion preparations in Figure 5.

The SPF (Sun Protection Factor) value is a test meter that explains the effectiveness of a product or substance which can act as a UV protector. The greater the SPF (Sun Protection Factor) value of a product or sunscreen-active substance, the more effective it will protect the skin. from the harmful influence of UV rays (Susanti *et al.* 2012.) Based on the results of calculating the SPF value in Figure 6. Graph of the combined SPF protection levels, Formula 1 (F1) with an extract composition of 100 mg sungkai and 100 mg cinnamon extracts obtained a high SPF value, namely 7.4 with the type of protection determined by the FDA (Food and Drug Administration) in Rizal & Maharani (2023) including the type of extra protection. Meanwhile, the results of the nanoemulsion preparation formula for the other two combination extracts, namely formulas F2 and F3, are included in the same type of protection, namely medium protection with an SPF (Sun Protection Factor) value for F2, 6.9 and F3, 6.7.

Furthermore, the results of the nanoemulsion preparation formula for cinnamon single extract, namely F4, have a minimum type of protection with an SPF value of 4, and the nanoemulsion preparation

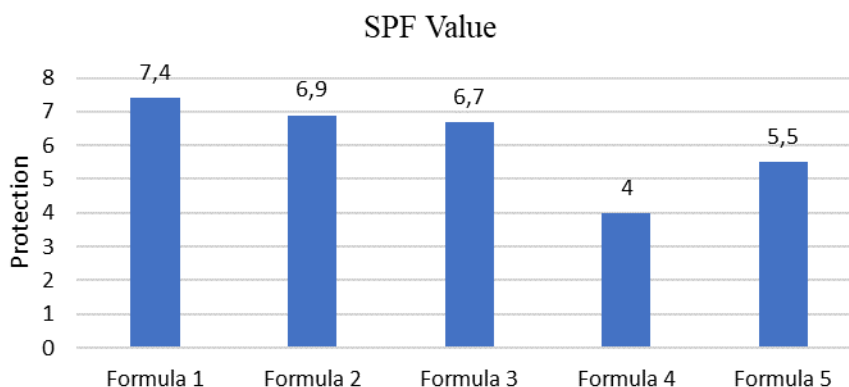


Figure 5. Combined graph of SPF protection levels

formula for single extract of sungkai, namely F5, has a medium type of protection with an SPF value of 5.5. From the results of the SPF value test in this study, it was concluded that the 3 combinations extract nanoemulsion preparation formulas were good protection preparations for the skin because they were able to provide an extra protective effect against direct exposure to sunlight. There was an increase in the SPF value of the preparations compared to the single formula of sungkai and cinnamon extracts. It is known that the combination of both sungkai and cinnamon extracts used as active substances has potential and meets the criteria for a sunscreen nanoemulsion preparation product in protecting the skin, overcoming the problem of free radical compounds in the body and inhibiting premature aging process.

The occurrence of differences in the SPF values produced in various combinations of extracts F1, F2, and F3 in the test can be seen that each nanoemulsion preparation formula has a different concentration in providing the active substance in the preparation with the same active substance weight, namely 200 mg. So that the optimal formula for the preparation as a sunscreen product is obtained. According to Ningsih & Atiqah (2020), the concentration of active substances in the nanoemulsion preparation formula will affect the results of the SPF (Sun Protection Factor) value. It is known that the more concentration of extract used, the higher the SPF value that will be obtained from the nanoemulsion preparation. Based on protection protection according to Suryadi et al., (2021), the SPF (Sun Protection Factor) value in the maximum category is that it can protect the skin from UV rays by inhibiting UV radiation by 93.3 – 95.9%, and for the ultra-category it can inhibit radiation. UV is 96.0 – 97.4%.

There is the ability of sunscreen activity from the nanoemulsion preparation of a combination of sungkai and cinnamon extract is known from several studies showing that it contains bioactive secondary metabolite compounds such as phenolics and

flavonoids as a source of strong antioxidants which have been proven to be able to inhibit radical compounds and skin problems such as premature aging and absorb light strongly. at UV light wavelengths. Phenolic compounds function in protecting plant tissue against damage due to solar radiation. Apart from that, flavonoid compounds in plants are also able to ward off ultraviolet (UV) induced radicals, and provide a protective effect against UV radiation by absorbing UV light. (Siampa *et al.* 2022; Antasionasti & Jayanto 2021; Fadlilaturrahmah *et al.* 2021).

Optimal Formula Irritation Test

The optimal formula for the nanoemulsion preparation of a combination of sungkai and cinnamon extracts applied to the volunteers' arms was observed. The results of the optimal formula irritation test are in Table 11.

Based on the results of the irritation test for the F1 nanoemulsion preparation in Table 11, the entire test group of 10 respondents were divided into three test groups with observation times of 24 hours, 48 hours and 72 hours, which were applied to the arm and showed no signs of irritation such as erythema and edema. The results of this test show that the optimal nanoemulsion preparation from a combination of sungkai and cinnamon extracts can be further used as a skin protection factor agent. The absence of irritation, such as erythema and edema on the skin applied to the volunteers' arms, means that the ingredients used in the nanoemulsion formulation are safe on the skin. Apart from that, the active substance used in the nanoemulsion preparation formula, a combination of sungkai and cinnamon extracts, is known to be able to inhibit the incidence of erythema and edema based on the results of research by Indarto *et al.* (2022) who reported that one of the common compounds used is the inhibition and absorption of UV light and is an antioxidant. Cinnamaldehyde from cinnamon can absorb UV radiation because it has a chromophore group from its aromatic core structure

Table 11. Optimal formula irritation test results

Respondent Group	Respondent's Name	Observation (hr)		
		24	48	72
(Teenager) 17 years – 22 years	A	-	-	-
	F	-	-	-
	P	-	-	-
	I	-	-	-
(Mature) 26 years – 35 years	E	-	-	-
	Y	-	-	-
	H	-	-	-
(Elderly) 50 years – 60 years	m	-	-	-
	D	-	-	-
	Y	-	-	-

Notes: (-): Does not cause erythema/edema; (+): Has caused erythema/edema

conjugated to a carbonyl group. As well as general compounds in the form of phenols and flavonoids in sungkai leaves as a source of antioxidants, they can function as photoprotection from UV radiation and can absorb UV-B rays.

CONCLUSION

Based on the results obtained from the research that has been carried out, it can be concluded that the optimal nanoemulsion formula is shown by formula 1 (F1) with an active ingredient concentration of 100 mg sungkai extract and 100 mg cinnamon extract. The characteristic results of the formula with a trasmitan percentage value of 98% have a clear, stable and transparent visual appearance, insoluble in non-polar solvents. hexane and ethyl acetate, and can dissolve in ethanol, methanol, water which are polar solvents, has a skin pH range of <4 and > 7, with an oil-in-water (o/w) emulsion type and a nanoemulsion droplet particle size of 35.0 nm with a polydispersity index (PI) value of less than 1 (0.479) so that the optimal formula F1 meets the requirements as a nanoemulsion product. The antioxidant activity of nanoemulsion in the optimal formula preparation (F1) has an IC50 value of 20.29 ppm, a very strong antioxidant level, preparation (F2) has an IC50 value of 40.7 ppm, a very strong antioxidant level, while the preparation (F3) has an IC50 value of 80, 83 ppm strong antioxidant level, preparation (F4) has an IC50 value of 90.5 ppm strong antioxidant level, and preparation (F5) has an IC50 value of 48.1 ppm very strong antioxidant level. The highest SPF activity value was obtained in the nanoemulsion preparation (F1) as the optimal formula with an SPF value of 7.4, which is included in the maximum protection according to the FDA (Food and Drug Administration), which is better compared to the four nanoemulsion preparation formulas made where (F2) has an SPF value of 6.9, (F3) with an SPF value of 6.7 and (F5) with an SPF value of 5.5 is included in medium protection. Meanwhile, (F4) has an SPF value of 4 which is included in the very weak protection of the formulation made

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